**C9orf72 DPR localization and effect of modifier genes on localization and protein levels.**

**a)** Cellular localization and expression of C9orf72 DPRs was evaluated by immunocytochemistry. DPRs were visualized using anti-FLAG antibody.

**b)** Upregulation of four karyopherin genes, KAP104, KAP114, KAP120 or KAP122, in yeast does not alter localization of (Pro-Arg)_50 DPR.

**c)** (Pro-Arg)_50 expression levels were evaluated in deletion strains that mitigate (Pro-Arg)_50 toxicity and under conditions of KAP104, KAP114, KAP120 and KAP122 overexpression. dhklΔ, gtr1Δ, sgo1Δ, ski8Δ, stp1Δ and uaf30Δ lowered levels of (Pro-Arg)_50.

**d)** Deletion of these 6 genes did not strongly lower levels of YFP expressed under the same promoter, indicating that their effect on (Pro-Arg)_50 levels was likely specific.
Effect of (Pro-Arg)$_{50}$ toxicity modifiers on TDP-43 and $\alpha$-synuclein ($\alpha$-syn) toxicity in yeast.

a) Gene deletions that suppressed (Pro-Arg)$_{50}$ toxicity were tested for unspecific effect on survival by introducing TDP-43 into these deletions. Only $ubr2\Delta$ and $ski8\Delta$ showed protective effect against TDP43 toxicity in yeast, while others did not. b) $ski8\Delta$ did not suppress $\alpha$-syn toxicity. $ubr2\Delta$ suppressed toxicity of all three proteins and therefore this deletion does not confer any specific protection against (Pro-Arg)$_{50}$ or TDP-43 toxicity, but might represent a common target for all three aggregation-prone proteins.
Supplementary Figure 3

KPNA3 upregulation does not affect localization of (Pro-Arg)_{50} in mouse primary cells.

Cells were transfected with (Pro-Arg)_{50} and GFP or KPNA3 (C-terminally V5 tagged). Neurons were stained with anti-FLAG antibody and anti-V5, were applicable. Representative images are shown.
Supplementary Figure 4

Effect of C9orf72 mutation on localization of proteins implicated in nucleocytoplasmic transport.

iNeurons from control subjects or C9orf72 mutation carriers were used to examine localization of a) LmnB and TNPO3, b) RanGAP1 and KPNA3, c) XPO5. We did not observe significant differences in the localization of these proteins in the control cells compared to C9orf72 mutation carrier cells.